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10/523,062	11/09/2005	Tamas Rujan	82371	8408

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EXAMINER	
PANDE, SUCHIRA	
ART UNIT	PAPER NUMBER
1637	
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/523,062	RUJAN ET AL.	
	Examiner	Art Unit	
	SUCHIRA PANDE	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *25 February 2008*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-52* is/are pending in the application.

4a) Of the above claim(s) *1-38,43-47 and 49-51* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *39-42,48 and 52* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date *3/11/2008*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Election/Restrictions

1. Applicant's election without traverse of group II invention (claims 39-51) in the reply filed on February 25, 2008 is acknowledged. Applicant has withdrawn claims 1-38, 43-47, 49-51; and added new claim 52.

Applicant has also elected species:

a. Species of nature of primer (claim 1 is generic)
i (primer molecules do not contain nucleic acid sequences complementary or identical to nucleic acid sequences of the target sequence which prior to treatment of step 2 contained a 5'-CG-3' site)—(claim 2);

b. Species of nucleic acid sample - xii (nucleic acid sample is comprised of human genomic DNA) (claim 9);

c. Species of numbers of primer pairs in a set- xiii (set is comprised of at least one but not more than 32 primer pairs) (claim 3);

d. Species of number of mismatches allowed - xxi (number of mismatches allowed for is one) (claim 23);

e. Species of number of nucleotides creating one gap allowed for in primer molecule - xxv (number of nucleotides creating one gap, when aligning the primer molecule sequence with the template sequence, allowed for, when virtually testing the amplification of unwanted products according to step 3 c) of claim 1 is less than 5% of the number of nucleotides of the primer molecule) (claim 19);

f. Species of nature of nucleic acid amplified by primer molecules - xxix (nucleic acid sequences that prior to treatment of step 2 comprised of more than two 5'-CG-3' sites) (claim 28);

g. Species of virtual PCR - xxxv (electronic PRC, taking as template nucleic acid the coding strand of the bisulfite converted human genome, the non-coding strand of the bisulfite converted human genome and both of the strands of the untreated human genome) (claim 31);

h. Species of measure of complexity- xxxvi (measure of complexity is a measure of linguistic complexity) (claim 42); and

i. Species of step carried out prior to performing step d) - xlii (excluding from the remaining primer pairs those pairs, which comprise of one primer molecule that in combination with another primer molecule in the set amplifies an unwanted product, when virtually testing according to step 3 c) under conditions allowing for a number of mismatching nucleotides of 20% of the number of nucleotides of the primer molecule) (claim 48).

Claims 39-42, 48 and 52 that are consonant with above elections will be examined in this action.

Information Disclosure Statement

2. The information disclosure statement (IDS) submitted on 3/11/2008 is in compliance with the provisions of 37 CFR 1.97. Accordingly, the information disclosure statement is being considered by the examiner.

Claim Rejections - 35 USC § 112

3. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

4. Claims 39-42, 48 and 52 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Regarding claim 39, currently the claim recites a method for designing primers according to claim 1. This recitation causes the claim to be indefinite because claim 1 is directed to method of amplification while claim 39 is directed to a method of designing primers. One of ordinary skill in the art can not fathom how can a method of designing primer depend of a method of amplification? This is because method of amplification can only be performed once the primers have been designed as the amplification method inherently requires primers.

Step d) of claim 39 refers to performing virtual PCR on treated and untreated nucleic acid templates and excluding those primers that amplify unwanted product. Claim does not specify what the treatment conditions are so one of ordinary skill is not clear as to what the meets and bounds of the claimed invention are.

Claims 40-42, 48 and 52 that depend from claim 1 or claim 39 share the same problem described above. Appropriate correction is required so one of ordinary skill clearly knows the meets and bounds of the claimed method of designing primers.

Claims 39-42, 48 and 52 all depend directly or indirectly from withdrawn claim 1, which is a method of amplification. Even though apparently the claims do not lack an antecedent basis and can be written in an independent form, but in principle they do have a problem. The problem being you can not conduct amplification without having the primers, so method of designing primers of claims 39-42, 48 and 52 can not depend on a method of amplification. Also the claims should be rewritten in an independent form so they do not depend from a withdrawn claim. Appropriate correction is required.

Claim 40 recites the limitation "adding the step of (e)" in the method according to claim 1. There is insufficient antecedent basis for this limitation in the claim. It is not clear to examiner where should step (e) be added when claim 1 does not recite a step (d) anywhere! Claim 1, as currently recited contains steps 1-4 with step 3 containing steps (a) through (c). Step (d) is not recited anywhere in the claim 1 hence step (e) lacks an antecedent basis i.e. it is not

clear after what stage (steps 1-4 of claim 1 this step (e) needs to be added. If it needs to be added to step 3 of claim 1, then step 3 is missing essential step 3(d) after which 3(e) can be added.

Regarding claim 52, the step d) of the recited claim is so convoluted that one of ordinary skill is left wondering what is actually claimed. Examiner has not considered this part at all for art purposes as it was not clear as to what kind of primer is being claimed.

Appropriate correction is required to address the indefinite aspects associated will all the above claims, so that it is clear to one of ordinary skill what are the meets and bounds of the claimed invention.

Claim interpretation

5. Due to the 112 2nd issues identified above for the purposes of searching prior art and its application, Examiner is assuming that applicant wishes to claim a method of designing primers of claim 39. Hence all the limitations in the claims directed to designing amplification primers under consideration that refer to claim 1 (method of amplification or steps that are part of method of amplification) will not be addressed in the rejections that follow.

Applicant has not defined complexity in the specification, Examiner is interpreting any primer that is used for PCR to have a specified level of complexity.

Step d) of claim 39 refers to performing virtual PCR on treated and untreated nucleic acid templates and excluding those primers that amplify unwanted product. Since claim does not specify what the treatment conditions

are, Examiner is interpreting any set of primers that prime the amplification of a PCR of correct molecular weight in e PCR on any desired template to mean that this set of primers meet the limitations specified in step d).

Claim Rejections - 35 USC § 103

6. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

7. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

8. Claims 39-41, 48 are rejected under 35 U.S.C. 103(a) as being unpatentable over Rozen and Skaletsky (2000) Bioinformatics methods and protocols/ edited by Stephen Misener and Stephen A. Krawetz. Humana Press Totowa NJ vol. 132: pp 365-386 in view of Schuler (1997) Genome methods vol. 7 pp 541-550 and Lexa et al. (2001) Bioinformatics vol. 17, no 2: pp 192-193.

Regarding claim 39, Rozen and Skaletsky teach a method for designing primers, comprising the steps of

a) selecting a pool of possible primer pairs per amplificate by means of a standard PCR primer design program using said nucleic acids as template (see whole article where Primer3 program is taught)

b) excluding those primer pairs which comprise of a primer that in combination with another primer molecule in the same set exceeds a threshold melting temperature (see page 372 where Rozen and Skaletsky teach options that specify which primers are acceptable. Among those options called constraints melting temperature referred to as primer Tm Min and Max, Max end stability and Max mispriming are taught. By this teaching the program clearly teaches excluding those primer pairs which comprise of a primer that in combination with another primer molecule in the same set exceeds a threshold melting temperature)

c) excluding those primer pairs which comprise of a primer that does not reach a specified level of complexity. As discussed above under claim interpretation any primer that is used for PCR will have a specified level of complexity. So by teaching a program to select desired primers for PCR Rozen and Skaletsky teach excluding those primer pairs which comprise of a primer that does not reach a specified level of complexity).

Regarding claim 41, Rozen and Skaletsky teach wherein said template nucleic acids are masked for repeats and SNPs before designing said primer molecules (see page 365 par. 1 introduction where a program that suggests PCR

primers for variety of applications are taught. By this broad teaching Rozen and Skaletsky teach wherein said template nucleic acids are masked for repeats and SNPs before designing said primer molecules) and wherein said standard PCR primer design program considers one or more of the following factors

length of amplificate, length of primer, melting temperature of the primers, dimer formation parameters, loop formation parameters (see page 365 par. 3 where each of these recited factors is listed under factors that are considered by Primer3 program), exclusion of unidentified or ambiguous nucleotides in the primer sequence (Rozen and Skaletsky teach the program considers the accuracy of a source sequence—by this teaching they teach exclusion of unidentified or ambiguous nucleotides in the primer sequence), exclusion of restriction enzyme recognition sites (Rozen and Skaletsky teach the program considers the primer location relative to particular regions of interest or to be avoided--by teaching the particular region to be avoided, Rozen and Skaletsky teach exclusion of restriction enzyme recognition sites).

Regarding claim 39, Rozen and Skaletsky, do not teach virtual testing of primers in electronic PCR.

step d) excluding those primer pairs which comprise of a primer that in combination with another primer molecule in the same set, under conditions allowing for one or more base mismatches per primer, amplifies an unwanted product when virtually tested using the treated and the untreated sample nucleic acid as template.

Regarding claim 39, Schuler teaches e PCR as a means for virtually testing PCR products obtained with primer pairs (see whole article). As explained under claim interpretation since the claim does not specify what the treatment conditions are, Examiner is interpreting any set of primers that prime the amplification of a PCR of correct molecular weight in e PCR to mean that this set of primers meet the limitations specified in step d). See abstract on page 541 of Schuler where they teach e PCR using a software tool to recover those unique sites in DNA sequences by searching for subsequences that closely match the PCR primers and have the correct order, orientation, and spacing that they could plausibly prime the amplification of a PCR product of the correct molecular weight. By teaching primers that result in the amplification of a PCR product of the correct molecular weight, Schuler teaches excluding those primer pairs which comprise of a primer that in combination with another primer molecule in the same set, under conditions allowing for one or more base mismatches per primer, amplifies an unwanted product when virtually tested on any desired nucleic acid as template).

Regarding claim 40, Schuler teaches adding the step of e) excluding from the remaining confirmed primer pairs those pairs which in said amplification step do not result in the amplification of the intended product when performing a single PCR experiment (See abstract on page 541 of Schuler where they teach e PCR using a software tool to recover those unique sites in DNA sequences by searching for subsequences that closely match the PCR primers and have the correct order, orientation, and spacing that they could plausibly prime the

amplification of a PCR product of the correct molecular weight. By teaching primers that result in the amplification of a PCR product of the correct molecular weight, Schuler teaches excluding from the remaining confirmed primer pairs those pairs which in said amplification step do not result in the amplification of the intended product when performing a single PCR experiment.)

Regarding claim 48, Schuler teaches excluding from the remaining primer pairs those pairs, which comprise of one primer molecule that in combination with another primer molecule in the set amplifies an unwanted product (see above as described for claim 39), when virtually testing under conditions allowing for a number of mismatching nucleotides of 20% of the number of nucleotides of the primer molecule (see page 543 where allowing up to N mismatching bases for each primer is taught---by this teaching Schuler teaches when virtually testing under conditions allowing for a number of mismatching nucleotides of 20% of the number of nucleotides of the primer molecule).

It would have been prima facie obvious to one of ordinary skill in the art to practice the method of Schuler in the method of Rozen and Skaletsky at the time the invention was made. The motivation to do so is provided to one of ordinary skill in the art by teachings of Lexa et al. and Schuler.

Lexa et al. teach virtual PCR and present an algorithm that uses sequence data from a public database to predict PCR products. They compare the output to real world PCR. They demonstrated the potential of the Virtual PCR (VPCR) by envisioning its uses in (i) evaluation of primers to be used in amplification from genomic DNA; and (ii) identification of PCR products with primers amplifying a

gene family (see page 192 section experimental verification). Based on the results they conclude the predictive ability of (VPCR) can improve by

"(i) using databases that cover the whole genome of a given organism;

(ii) identification of PCR products across boundaries of GenBank entries;

(iii) replacement of BLAST routines with calculations of primer binding; and

(iv)---- With these improvements the algorithm could become a standard component of primer design software."

Schuler state "one straightforward application of e-PCR is the large scale assignment of sequence database record to map positions.----To simplify the process and make it more widely available, an e-PCR search facility recently has been added to the NCBI site on the World Wide Web.----When developing new markers for mapping studies, e-PCR can be used to test potential primers in various ways before actually incurring the expense of oligonucleotide synthesis" (see page 548 par. 2-4).

Thus providing reasonable expectation of success that virtual testing methods can be applied to design and test potential primers in various ways depending on the desire of the person conducting the test and the final application for which these primers are being designed. This could be done for large scale projects and could result in significant savings both in terms of cost and time in which a project may be successfully accomplished.

9. Claim 42 and 52 are rejected under 35 U.S.C. 103(a) as being unpatentable over Rozen and Skaletsky; Schuler and Lexa et al. as applied to

claim 39 above further in view of Bolshoy et al. (1997) Nucleic acids res. Vol.25 No 16 pp 3248-3254 and Laird et al. (US pat. 6,331,393 B1 issued December 18, 2001).

Regarding claim 41, Rozen and Skaletsky; Schuler and Lexa et al. teach method of claim 39, but do not teach wherein said measure of complexity is a measure of linguistic complexity.

Regarding claim 41, Bolshoy et al. teach wherein said measure of complexity is a measure of linguistic complexity (see page 3249 section Linguistic complexity where program for calculation of linguistic complexity are taught).

Regarding claim 52, Rozen and Skaletsky; Schuler and Lexa et al. as applied to claim 39 teach steps a) through c) and step e) of instant claim.

Regarding step d) of claim 52, Laird et al. teach regions containing CpG site.

Regarding step d) of claim 52, Bolshoy et al. teaches linguistic complexity determination program for determining regions containing the requisite no of CpG sites.

Regarding step d) of claim 52, Schuler teaches excluding primer pairs that give unwanted amplification products by virtual testing. Rest of step d) is not being considered as Examiner could not discern what is actually claimed.

It would have been prima facie obvious to one of ordinary skill in the art to practice the method of Bolshoy et al. in the method of Rozen and Skaletsky;

Schuler and Lexa et al. at the time the invention was made. The motivation to do so is provided to one of ordinary skill by teachings of Bolshoy et al.

Bolshoy et al. state "The complexity discrimination approach proposed here allows *a priori* identification of the sequences more likely to have signal-to-noise ratios, by calculating the complexities of all the sequences in the collection (e.g. by the measure of linguistic complexity, as in this work)an removing or giving less weight to those that are most complex. We believe that this approach will prove useful in the extraction of other degenerate biological signals----. This study provides only a single illustration of potentially important approach utilizing sequence complexity for the analysis of overlapping degenerate patterns." See page 3253 last par.).

In view of the above explicit teaching and by the knowledge of molecular biology one of ordinary skill knows that in higher eukaryotes DNA is methylated only at cytosines located 5' to guanosine in the CpG dinucleotide. This modification has important regulatory effects on gene expression predominantly when it involves CpG rich areas (CpG islands) located in the promoter region of a gene sequence. Extensive methylation or changes in methylation pattern are associated with altered gene expression and tumor formation (See Laird et al. col. 1 lines 14-27). Thus one of ordinary skill recognized CpG islands that may be methylated or not as regions containing overlapping degenerate patterns.

Hence once of ordinary skill has a reasonable expectation of success in determining the linguistic complexity of the CpG containing regions that are spread over the genome of a normal vs cancer patient. By combining the

linguistic complexity of Bolshoy et al. as applicable to the regions containing CpG islands in the samples from normal and cancer patients and further using the method of designing primers taught by Rozen and Skaletsky; Schuler and Lexa et al.; one or ordinary skill in designing primers that will be able to analyze the PCR products from the two types of templates (normal vs cancer DNA) and draw conclusions regarding the sets of genes whose altered expression may contribute to the disease onset and progression.

Conclusion

10. All claims 39-42, 48 and 52 under consideration are rejected.

11. Any inquiry concerning this communication or earlier communications from the examiner should be directed to SUCHIRA PANDE whose telephone number is (571)272-9052. The examiner can normally be reached on 8:30 am -5:00 pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 571-272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

Suchira Pande
Examiner
Art Unit 1637

/Teresa E Strzelecka/
Primary Examiner, Art Unit 1637

May 5, 2008